

WHAT IS CLAIMED IS:

1. An automatic focusing method comprising:

scanning light from a light source which passes a
confocal pattern on a sample through an objective lens
while relatively moving one of the sample and the
objective lens along a direction of an optical axis;

acquiring two or more sectioning images by
converting the light from the sample which penetrates
the confocal pattern through the objective lens by
photoelectric conversion means; and

changing an opening diameter of the variable
diaphragm arranged at the pupil position of the
objective lens or a conjugated position to the pupil
position thereof to reduce a NA of the objective lens
when focusing is not obtained and repeating an
operation of taking two or more sectioning images by
the photoelectric conversion means and obtaining the
focusing position.

2. An automatic focusing method comprising:

scanning the sample with light from a light source
which passed a confocal pattern while moving one of a
sample and an objective lens along the direction of an
optical axis at a predetermined sampling interval;

acquiring two or more sectioning images by
converting light from the sample which penetrates the
confocal pattern through the objective lens by the
photoelectric conversion means;

obtaining a focusing position according to a predetermined function based on the plurality of sectioning images taken by the photoelectric conversion means; and

5 changing an opening diameter of the variable diaphragm arranged at the pupil position of the objective lens or a conjugated position to the pupil position thereof to reduce a NA of the objective lens when focusing is not obtained and repeating an
10 operation of taking two or more sectioning images by the photoelectric conversion means and obtaining the focusing position.

3. The automatic focusing method according to claim 2, wherein an objective lens with low
15 magnification and high NA is used for the objective lens.

4. The automatic focusing method according to claim 2, wherein two or more sectioning images are taken without changing the predetermined sampling
20 interval when the NA of the objective lens is changed.

5. The automatic focusing method according to claim 2, wherein an operation to which the focusing position is obtained is repeated until three or more sectioning images are acquired.

25 6. The automatic focusing method according to claim 2, wherein whether the sectioning image uses data of a part where disorder is caused by an aberration of

the objective lens is judged and the sectioning image is acquired by reducing the NA of the objective lens when the disordered data is used.